IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re the Application of)
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ROGER P EKINS)
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Serial No: 08/447,820)
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Filed: 23 May 1995)
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For: DETERMINATION OF AMBIENT CONCENTRATIONS OF SEVERAL ANALYTES

Examiner: M Woodward

Group Art Unit: 1815

#13
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3/1/98

DECLARATION OF ROGER PHILIP EKINS

I, Professor Roger Philip Ekins, declare as follows:

1. I am the sole inventor of the above-identified patent application. At the present time, I am Professor of Molecular Endocrinology at University College London Medical School. My curriculum vitae accompanies this declaration.

2. I have read the Final Rejection of 2 September 1997 and the Advisory Action of 10 December 1997 from the US Patent Office, WO84/01031 (Ekins '031), US Patent No 4,385,126 (Chen et al '126) and the declaration of Dr Johann Berger. I comment on the Examiner's rejection based on my earlier patent application WO84/01031 (Ekins '031) as follows.

3. Ekins '031 relates to the development of assays which are sample-volume independent, that is it shows that by using a small amount of binding agent (e.g. antibody), one may develop assays in which the fractional occupancy of the binder is independent of the binder concentration (described in Ekins '031 as "ambient analyte" conditions). However, it is important to realise that Ekins '031, and in particular the equations set out in this application, do not relate to assay sensitivity, nor do they in any way suggest that use of a small amount of binding agent could - under certain circumstances - enhance sensitivity.

4. Thus, it does not follow that assays designed in the manner disclosed in Ekins '031 could be optimised to arrive at the invention claimed in the above mentioned application, nor would Ekins '031 lead a person skilled in the art to believe that, by optimising such assays, higher sensitivity could be obtained. Thus, I disagree with the Examiner that "given the equations of Ekins one would proceed to optimize the assay as one saw fit", and so arrive at the presently claimed invention. On the contrary, the invention in the above mentioned application relates to providing highly sensitive assays by immobilising amounts of binding agent less that 0.1V/K moles at high density, e.g. as microspots. That this approach enhances sensitivity is unexpected and not predictable from the earlier and distinct invention set out in Ekins '031.

5. It should be emphasised that contemporary opinion in the field held that to maximise assay sensitivity, considerably larger amounts of binding agent than those used in Ekins '031 were optimal in the case of "competitive" assays (typically to bind 33% or 50%of the analyte present in a sample (ie 0.5/K - 1/K)), and even larger amounts of binding agent (ie >>1/K - 10/K) were optimal in the case of "non-competitive" assays (typically to bind all or nearly all of the analyte in a sample). There is no reason to suppose that Ekins '031 would overturn these views since it relates solely to the design of assays which are sample-volume independent and does not address the issue of sensitivity. In other words, a person of ordinary skill in the art would inevitably conclude that an assay designed in accordance with Ekins '031 to be independent of sample volume would concomitantly suffer a considerable loss in sensitivity. Nothing in the equations in this application would lead the person of ordinary skill in the art to think otherwise, nor point to the way in which the loss of sensitivity thought to be an inevitable consequence of the use of a "small" amount of binding agent might be overcome.

6. These considerations underlie and explain Dr Berger's initial total disbelief mentioned in his declaration (which I personally witnessed and emphatically confirm) that an assay designed in accordance with the instant specification would yield an assay system capable of measuring even lower analyte concentrations than conventional methodologies.

7. The Examiner attempts to repudiates Dr Berger's comments on the Chen et al '126 references stating that "in order to employ the Ekins assay one must know the amount of binding agent present". In the sense implied by the Examiner this is wholly untrue. In fact the use of dual labelling in accordance with the instant specification relieves the assay manufacturer of having to know or to keep constant the exact amount of binding agent affixed to a support. That is because in practising the assay of the instant specification one simply needs to know that the total amount of binding agent present is less than the threshold level of 0.1V/K moles, where K is the effective affinity constant of the binding agent, rather than the exact amount of binding agent present.

8. This is quite contrary to the teaching in Chen et al '126, where the use of a label attached to the "capture" binding agent (i.e. that located on the solid phase) is explicitly intended to confirm that the standard amount of capture binding agent is present (i.e. it serves a quality control function). In an assay performed in accordance with the instant specification it is unnecessary to determine how much binding agent is present, and it is therefore unnecessary (inter alia) to scan the entire area on which the binding agent is deposited, see the application at page 11, lines 18-23. Moreover manufacturers frequently encounter difficulties in ensuring that the same amount of binding agent is present in every incubation tube. Thus, it is a distinct advantage of the present invention that one does not need to know the precise amount of binding agent present.

9. In summary, the invention described in Ekins '031 discloses an assay system that yields results independent of sample volume. Ekins '031 in no way indicates that contemporary teachings in 1987 relating to the design of high sensitivity assays were wrong. Thus, there is no reason based on Ekins '031 to suppose that those of ordinary skill in the art would re-examine the results of their past optimization experiments or that any further

experiments of this nature would reverse conclusions widely accepted in the art regarding the disadvantages of the use of low binding agent concentrations in regard to sensitivity.

10. In contrast, the invention of the above mentioned patent application represents a quite different approach to assay design and one which permits the construction of miniaturized microarrays. In this invention, the use of a small amount of binding agent surprisingly contributes to the high sensitivity achieved, and is a component of the invention in combination with immobilising the binding agent at high density, e.g. as a microspot, to achieve sensitivities no less (or even much higher than) those achievable using conventional assay designs. This is not derivable from Ekins '031, much less as an inevitable result of routine assay optimisation.

I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true and further that these statements were made with the knowledge that wilful false statements and the like so made are punishable by fine or imprisonment, or both, under §1001, Title 18 of the United States Code, and that such wilful false statements may jeopardize the validity of this application or any patent issued thereon.

Date: Feb 24th 1998 Signed: Ralph Ekins